

**EGG PREDATION RISK TRIGGER ADULT HOVERFLY (DIPTERA: SYRPHIDAE)
TO AVOID LAYING EGGS IN PATCHES ATTENDED BY LADYBIRD LARVAE
(COLEOPTERA: COCCINELLIDAE)**

**RESIKO PREDASI PADA TELUR MEMICU LALAT APUNG (DIPTERA:
SYRPHIDAE) UNTUK MENGHINDARI PELETAKAN TELUR PADA TEMPAT
YANG DIHADIRI OLEH LARVA KUMBANG KOKSI
(COLEOPTERA: COCCINELLIDAE)**

Nugroho Susetya Putra

*Faculty of Agriculture, Gadjah Mada University
Sekip I Bulaksumur, Yogyakarta 55281, Indonesia*

Hironori Yasuda

*Faculty of Agriculture, Yamagata University
Wakaba-machi 1-23, Tsuruoka 997-8555, Yamagata, Japan*

INTISARI

Preferensi peletakan telur oleh lalat apung predator, *Episyrphus balteatus* pada koloni kutu aphid yang dihadiri oleh predator potensial, yaitu larva tiga spesies kumbang koksi predator yang menimbulkan dampak serius pada telur diamati di laboratorium menggunakan uji tanpa pilihan. Hasil uji menunjukkan bahwa spesies kumbang koksi yang paling besar dan agresif, *Harmonia axyridis* menimbulkan dampak negatif paling serius dengan menyerang dan memangsa telur lalat apung dibandingkan dua spesies kumbang koksi yang lain. Spesies dan tahap perkembangan larva kumbang koksi juga menentukan tingkatan resiko predasi pada telur lalat apung. Pemilihan tempat peneluran oleh lalat apung kemudian dikorelasikan dengan tingkat predasi yang disebabkan oleh larva kumbang koksi pada telur lalat apung. Hasil pengujian menunjukkan bahwa lalat apung akan meletakkan telur dalam jumlah paling sedikit pada koloni yang dihadiri oleh kompetitor terkuat, yaitu larva *H. axyridis*, dan meletakkan telur dalam jumlah paling banyak pada koloni yang dihadiri oleh kompetitor terlemah, yaitu *Scymnus posticalis*. Sebagai tambahan, dampak yang ditimbulkan oleh larva kumbang koksi instar keempat lebih kuat dibandingkan larva kumbang instar pertama.

Kata kunci : Predasi, Peletakan telur, Larva kumbang koksi

ABSTRACT

Oviposition preference of a predatory hoverfly, *Episyrphus balteatus* on the presence of its potential predators, the ladybird larvae which are inflicted serious impacts on its eggs was examined in a non-choice test. Our results revealed that the biggest and the most aggressive species of ladybird, *Harmonia axyridis* caused the worst impact on hoverfly eggs by attacking and feeding on. The species and developmental stages of ladybird were attributed to the level of predation risk. We correlated the oviposition site selection by hoverfly females to the egg predation risk level inflicted by ladybird larvae. Hoverfly females laid the least number of eggs on the patches attended by the strongest competitor, the larva of *H. axyridis*, and tended to lay the highest number of eggs on colonies attended by the weakest competitor, the larva of *Scymnus posticalis*. In addition, the impact of the fourth instar larva of ladybirds was stronger than of the first instar larva.

Key words : Predation, Laying eggs, Ladybird larvae

INTRODUCTION

Oviposition behavior of predatory arthropods is not only guided by the performance of prey (i.e. availability and quality), but is also influenced by the presence of potential predators or competitors. Some studies have revealed that in general, the presence of competitors repels the preference of females to visit oviposition sites and to lay their eggs (Blaustein et al., 2004), or at least reduces the number of eggs laid (Taylor et al., 1998; Takizawa et al., 2000). Thus, the trade-off mechanism is applied by females to adjust a fit between gaining an optimum prey availability and avoiding the risks from predation by the stronger predators as commonly shown in non-predatory insects (e.g. Gross & Price, 1998; Mira & Bernays, 2002). However, there are only little studies have been clarified such mechanism on predatory insects.

It is well known that the oviposition site selection by females often faces high risk due to predation by within-species (i.e. cannibalism) or inter-species of individuals (Schellhorn & Andow, 1999; Kiflawi et al., 2003; Eitam & Blaustein, 2004), hence finding enemy-free space is likely to be applied by both herbivorous and carnivorous arthropods to reduce predation risks by adjusting the choice of oviposition sites (Mira & Bernays, 2002; Ballabeni et al., 2001; Singer et al., 2004). For example, the sphingid moth *Manduca sexta* tended to lay eggs on a non-solanaceous host, *Proboscidea parviflora* which is actually a low-quality food than a high-quality food, a solanaceous host, *Datura wrightii* for reducing the predation risk by parasitism (Mira & Bernays, 2002).

Detrimental impacts of ladybird larvae on other species of aphidophagous insects have been proven in various studies (e.g. Ferguson & Stiling, 1996; Lucas et al., 1998; Felix & Soares, 2004), since they coexist in the same niche. Ladybird larvae often tend to be superior as an intraguild predator by reducing the

survivorship of the inferior species (Yasuda & Ohnuma, 1999; Hindayana et al., 2000; Meyhofer & Hindayana, 2000). In addition, hoverfly larvae may be vulnerable to being predated by potential intraguild predator such as ladybird larvae due to their low mobility. For instance, ladybird larva inflicted serious impacts on hoverfly larvae, *Episyrphus balteatus* de Geer by attacking and consuming the larvae (Hindayana et al., 2000). This study emerges an assumption that predatory hoverfly would consider to select oviposition sites which are free from the attendance of potential predators besides finding growing colonies of aphids as the most suitable sites (Kan & Sasakawa, 1986; Kan, 1988a; Kan, 1988b).

A couple of laboratory experiment was conducted to understand the oviposition behavior of predatory hoverfly, *E. balteatus* as a consequence of predation risk on its eggs by larvae of three species of ladybirds, *Harmonia axyridis* Pallas, *Propylea japonica* Thunberg, and *Scymnus posticalis* Sicard. Out of three ladybird species, *H. axyridis* is the biggest and the most aggressive species that consume not only on aphids, but also other arthropods such as other ladybird species and spiders (Yasuda & Shinya, 1997; Phoofole & Obrycki, 1998; Yasuda & Ohnuma, 1999; Sato & Dixon, 2004), while *S. posticalis* is the smallest and less mobile species (Agarwala & Yasuda, 2001), and *P. japonica* is intermediate in body size between those two species (Putra, personal observation). The differences in body size, voraciousness, and food habit of ladybirds are suggested may function on the strength of predation on hoverfly eggs. Furthermore, *E. balteatus* is described as a voracious and a polyphagous species (Owen & Gilbert, 1989; Gilbert & Owen, 1990; Tenhumberg, 1995).

Predation by the first and fourth instars of ladybird larvae on hoverfly eggs was examined firstly. We assumed that the level of egg predation would be determined by the developmental stages and biological traits (i.e.

body size, aggressiveness, food habit) of ladybird larvae. Thus, the fourth instar of larva would inflict the worse impact on hoverfly's eggs than the first instar. Secondly, the adjustment of oviposition site selection by females as a response of predation risk on their eggs was then examined. We predicted that the level of oviposition response by hoverfly females was agreed with the level of predation on their eggs.

MATERIALS AND METHODS

Insect rearing. Adult of hoverfly, *E. balteatus* and three species of ladybirds, *H. axyridis*, *P. japonica*, and *S. posticalis* were collected from Yamagata University farm and Akagawa Riverbank, at Tsuruoka, Yamagata Prefecture, Japan. Same numbers of females and males were then reared in muslin cage (60 x 60 x 70) cm³ with 20-30 individuals in total. Food for adults was provided by putting crushed-pollen in a small Petri dish (5 cm in diameter and 1 cm in height). In addition, for providing water and honey for adults, an aluminum stick with thin layer of honey at the upper part and with kitchen paper embedded at a half of the lower part (40 cm in length and 2 mm in diameter) was inserted into a 500 ml plastic bottle filled with water through a small hole punctured in a cap. When some females and males have seen in pairs, a couple of cutting bean plants with *Aphis craccivora* Koch colonies were introduced into the cage as oviposition sites. Eggs were collected daily, and then were placed into a Petri dish for materials. We used eggs which are laid by females on less than 12 hours for study. A pair of adult ladybird was placed in a Petri dish (5 cm in diameter and 1 cm in height) and fed with *Acyrtosiphon pisum* (Harris) for *H. axyridis* and *P. japonica*, while *A. craccivora* for *S. posticalis*. All insects were maintained in a controlled room at 23°C, 75% of relative humidity, and under a photoperiod of 16L: 8D.

Egg predation by ladybird larvae. To obtain ladybird larvae for examination, new individuals were reared on a plenty number of aphid in a small Petri dish (5 cm in diameter and 1 cm in height) as mentioned in the insect rearing method. The first instar larvae was obtained on less than 12 hours after they hatched, while the fourth instar larvae were obtained within 24 hours after molting. These individuals were starved for 6 hours prior to the test in order to increase the hunger level. Twenty individuals of eggs were arranged in two rows in the inner base of Petri dish prior to release an individual of ladybird larva. A small piece of hard-paper with the blue color was placed in the outer base of Petri dish to make a contrast view with the eggs for facilitating observation. The test was conducted for 10 minutes for each individual of ladybird species with 20 replications per combination of species and developmental stages. The number of eggs consumed by ladybird larvae were counted and was stated as the percent of egg predated by using a formula as follow: (number of eggs consumed-20) x100. Two-way ANOVA was performed to determine the impacts of ladybird species and larval stages on the percent of egg predation (Sokal & Rohlf, 1995).

Effect of the attendance of ladybird larvae on the oviposition response of adult hoverfly.

The effect of a ladybird attendance on the oviposition response of adult hoverfly was examined by a non-choice test using 30 different gravid females for each species with the same age, i.e. 25 - 30 days after eclosion. A cutting bean plant (20 cm in height) which has been infested with 20 - 25 individuals of apterae of *A. craccivora* at the apical portion was put into a bottle (5 cm in height) with water, and it was covered with a cylinder plastic cage (8 cm in diameter and 42 cm in height) as an oviposition site. One gravid female was released into one of seven treatments as mentioned as follow: (1) an aphid colony

without any ladybird larva as control, (2) aphid colonies with a fresh-frozen ladybird larva (i) a first larva or (ii) a fourth larva of *H. axyridis*, (iii) a first or (iv) a fourth instar larvae of *P. japonica*, and (v) a first or (vi) a fourth instar larvae of *S. posticalis*. Thus, seven treatments were applied for seven hours for each gravid female by following random orders. The number of eggs laid on the bean plant at each treatment was counted. One-way ANOVA was performed to find significant differences among treatments, and means were compared by post-hoc Fisher LSD (Sokal & Rohlf, 1995). Furthermore, to determine a correlation between the effects of egg predation by ladybird larvae and the oviposition preference of hoverfly females, the Pearson's Correlation test was used (Sokal & Rohlf, 1995).

RESULTS

Egg predation by ladybird larvae. Our results revealed that *H. axyridis* larva inflicted the highest risk of egg predation, while the *S. posticalis* caused the least risk (Fig. 1). In addition, the effect of the fourth instar larva was stronger than of the first instar larva. The statistical analysis showed that the effect of ladybird species and developmental stages were significantly different (for ladybird species, $F_{(2,114)} = 67.18$, $p < 0.001$; for developmental stages, $F_{(1,114)} = 357.31$, $p < 0.001$; for interaction of ladybird species and developmental stages, $F_{(2,114)} = 38.71$, $p < 0.001$).

Effect of the attendance of ladybird larvae on the oviposition response of adult hoverfly. The response of *E. balteatus* females on aphid colonies attended by the first and fourth instar larva of three ladybird species was varied significantly among treatments (Fig. 2, $F_{(6,203)} = 3.55$, $p < 0.05$). In general, the females tended to lay less number of eggs on aphid colonies with a ladybird larva than without a ladybird

larva. Furthermore, it seems that the fourth instar larva of ladybirds gave the stronger influence than the first instar larva to repel females for laying eggs, although there were only significant differences in the number of eggs laid between the control and the treatments with the fourth instar larva of *H. axyridis* and *P. japonica*. In overall, negative impacts of the fourth instar larva of the ladybirds on the number of eggs laid by the females were as follows: *H. axyridis* > *P. japonica* > *S. posticalis*.

Egg predation-oviposition preference correlation. The result showed that egg predation risks by ladybird larvae reduced the number of eggs laid by female hoverflies. Thus, the higher percent of egg predation was followed by the decreasing number of eggs laid ($r = -0.27$, $n = 140$, $p < 0.01$). This fact supported a prediction on the significant effect of the presence of competitor, i.e. ladybird larva, on the decrement of oviposition preference by female hoverfly (Fig. 3).

DISCUSSION

Egg predation. Present study revealed that the strength of ladybird predation on hoverfly's eggs was differed in the species and developmental stages involved. The biggest species, *H. axyridis* larva caused the worst level of predation on hoverfly eggs than the other two species by consuming more eggs in the given time. In addition, generally, the fourth instar larva of ladybird inflicted the stronger impact than the first instar larva.

Egg predation by within- or inter-specific individuals is well known in aphidophagous guild (e.g. Schellhorn & Andow, 1999; Gagne et al., 2002). Biological characteristics such as body size, voraciousness, and aggressiveness of predator may lead to the strength of predation on inferior species (Lucas et al., 1998; Snyder & Wise, 1999;

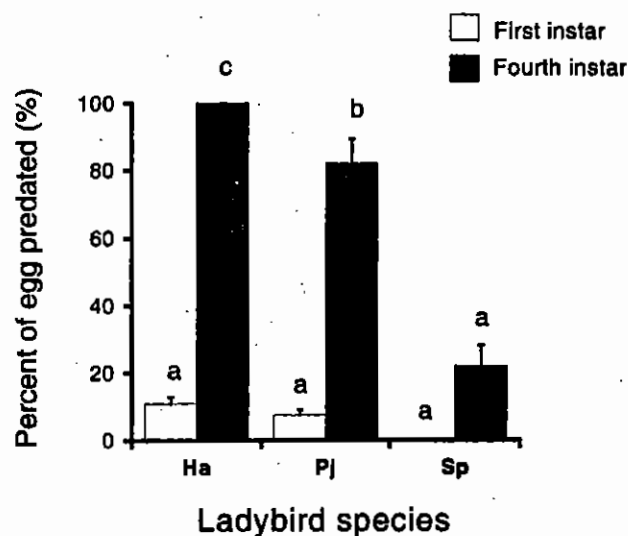


Fig. 1. Percent of predation on hoverfly eggs by the first and fourth instar larvae of three ladybird species, *Harmonia axyridis* (Ha), *Propylea japonica* (Pj), *Scymnus posticalis* (Sp). Significant difference among treatments is indicated by different alphabets ($p < 0.05$; Fisher-LSD post hoc test).

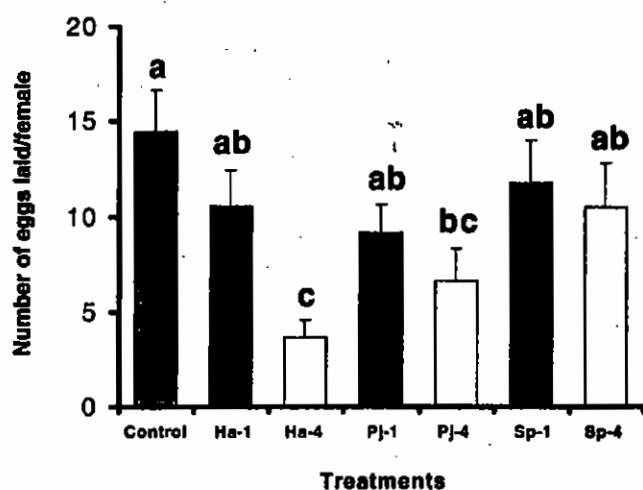


Fig. 2. Number of eggs laid (Mean \pm SE) by the hoverfly, *E. balteatus* as a response to the presence of ladybird larvae, *H. axyridis* (Ha), *P. japonica* (Pj), *S. posticalis* (Sp). The numeral 1 and 4, and control show the treatments with first, fourth instar larva, and without a larva, respectively. Different alphabets above bars indicated significantly differences among treatments ($p < 0.05$; Fisher-LSD post hoc test).

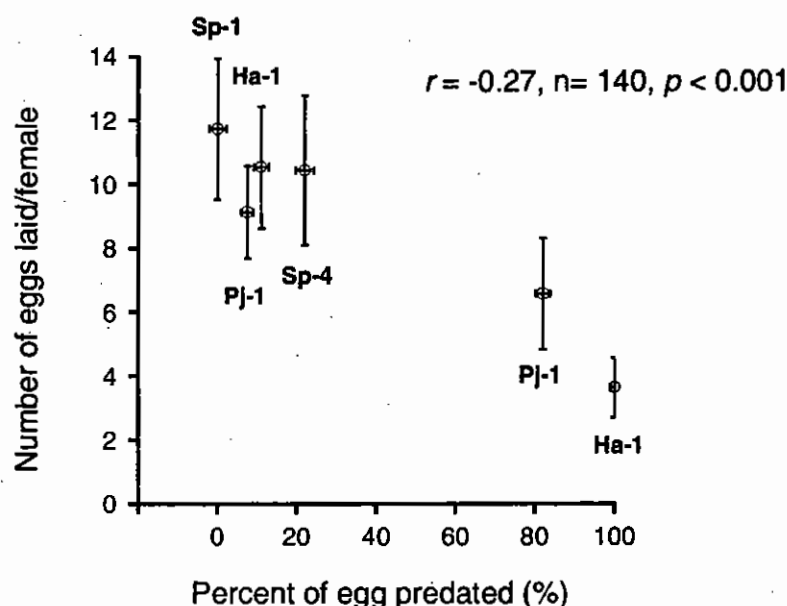


Fig. 3. Relationship between percentage of predation (%) on hoverfly eggs and number of eggs laid by hoverfly females on the presence of ladybirds, *H. axyridis* (Ha), *P. japonica* (Pj), and *S. posticalis* (Sp). The numeral 1 and 4 show the treatments with first and fourth instar larva, respectively.

Hindayana *et al.*, 2000), in addition to the fact that the body size is noticed as the most important factor determining predation levels (Felix & Soares, 2004). Our result showed that out of the three species, *H. axyridis*, especially of the fourth instar larva has the biggest of body size compared to the other two species.

Besides to body size effect, we suggested that the highly aggressiveness of *H. axyridis*, the fourth instar in particular was accounted to the high detrimental impacts on other species as also revealed in other studies (Brown & Miller, 1998; Yasuda & Ohnuma, 1999). This phenomenon was revealed in our study by highly significant destruction impact of *H. axyridis* larva on hoverfly eggs. During ten minutes of test, the destruction of larvae on eggs was always reaching a hundred percent (Fig. 1).

However, we suggested that the strength of impacts might be reduced in natural condition due to the availability of preys in fields. In addition, although egg was described as the weakest stage in the development of arthropod, some species develop chemical protection on their egg's surface to repel predation as revealed in predatory coccinellid (e.g. Omkar *et al.*, 2004). Our results were then emerged a suggestion on the different impacts of ladybirds, besides the biological traits of ladybirds (i.e. body size and aggressiveness). The higher impacts of *H. axyridis* larva than other of two species on hoverfly's eggs might be due to the higher acceptability of this species on the hoverfly egg, although so far, there are no studies revealing this assumption.

Effect of the attendance of ladybird larvae on the oviposition response of adult hoverfly.

Oviposition response of *E. balteatus* females correlated to the effect of ladybird larvae on their eggs. Thus, the females tended to lay a lesser number of eggs on aphid colonies attended by ladybird larvae which were causing the worse effects on their eggs.

It is well known that prey quantity and quality strongly influence the oviposition preference of hoverfly, since the females tend to choose the new colonies of aphid (Kan & Sasakawa, 1986; Kan, 1988a, b) in addition to consider the aphid quality (Sadeghi & Gilbert, 2000).

In our results, the females also seemed to consider the presence of ladybird larva which is potentially triggering risks on their offspring as also revealed in phytophagous arthropods (e.g. Mira & Bernays, 2002; Kanno & Harris, 2002). It is well known that arthropods applied some strategies to reduce the risks from being predated by selecting enemy free patches for laying their eggs (Gross & Price, 1988; Denno et al., 1990; Novotny et al., 1999; Singer et al., 2004).

In addition with the facts that female hoverfly tended to lay their eggs on newly aphid colonies, a more complex behavior of oviposition preference might also be applied to adjust a fit between larval performance and oviposition preference as shown in other arthropods. For instance, in order to reduce the predation risk by the parasitoid, in nature, the larva of generalist arctiid, *Estigmene acraea* preferred to consume a low quality host, *Senecio longilobus* which contains an alkaloid (Singer et al., 2004). This study indicated that the value of enemy-free space supersedes the value of food quality. Therefore, we suggested that the presence of potential predators might have negative consequences on the finding of optimal food in hoverfly, which in turn might also reduce the abundance of hoverfly in patches which are occupied by the stronger competitors.

The study to prove such assumption should be done to provide a better understanding on the structuring of arthropod community.

REFERENCES

Agarwala, B.K. & H. Yasuda. 2001. Larval interactions in aphidophagous predators: effectiveness of wax cover as defense shield of *Scymnus* larvae against predation from syrphids. *Entomol. Exp. Appl.* 100: 101-107.

Ballabeni, P., M. Włodarczyk & M. Rahier. 2001. Does enemy-free space for eggs contribute to a leaf beetle's oviposition preference for a nutritionally inferior host plant? *Functional Ecol.* 15: 318-324.

Blaustein, L., M. Kiflawi, A. Eitam, M. Mangel & J.E. Cohen. 2004. Oviposition habitat selection in response to risk of predation in temporary pools: mode of detection and consistency across experimental venue. *Oecologia* 138: 300-305.

Dal., P. Drozd, M. Kasbal, B. Isua, R. Kutil, M. Manumbor & K. Molem. 1999. Predation risk for herbivorous insects on tropical vegetation: a search for enemy-free space and time. *Aus. J. Ecol.* 24: 477-483.

Omkar, A. Pervez & A.K. Gupta. 2004. Role of surface chemicals in egg cannibalism and intraguild predation by neonates of two aphidophagous ladybirds, *Propylea dissecta* and *Coccinella transversalis*. *J. Appl. Entomol.* 128: 691-695.

Owen, J. & F.S. Gilbert. 1989. On the abundance of hoverflies (Syrphidae). *Oikos* 55: 183-193.

Phoofolo, M.W. & J.J. Obrycki. 1998. Potential for intraguild predation and competition among predatory Coccinellidae

and Chrysopidae.

Brown, M.W. & S.S. Miller. 1998. Coccinellidae (Coleoptera) in apple orchards of eastern West Virginia and the impact of invasion by *Harmonia axyridis*. *Entomol. News* 109(2), 136-142.

Denno, R.F., S. Larrison & K.L. Olmstead. 1990. Role of enemy-free space and plant quality in host-plant selection by willow beetles. *Ecology* 71: 124-137.

Eitam, A. & L. Blaustein. 2004. Oviposition habitat selection by mosquito in response to predator (*Notonecta maculata*) density. *Physiol. Entomol.* 29: 188-191.

Felix, S. & A. O. Soares. 2004. Intraguild predation between the aphidophagous ladybird beetles *Harmonia axyridis* and *Coccinella undecimpunctata* (Coleoptera: Coccinellidae): the role of body weight. *Euro. J. Entomol.* 101: 237-242.

Ferguson, K.I. & P. Stilling. 1996. Non-additive effects of multiple natural enemies on aphid populations, *Oecologia* 108: 375-379.

Gagne, I., D. Coderre & Y. Mauffette. 2002. Egg cannibalism by *Coleomegilla maculata* lengi neonates: preference even in the presence of essential prey. *Ecol. Entomol.* 27: 285-291.

Gilbert, F.S. & J. Owen. 1990. Size, shape, competition, and community structure in hoverflies (Diptera: Syrphidae). *J. Anim. Ecol.* 59: 21-39.

Gross, P. & P.W. Price. 1988. Plant influences on parasitism of two leafminers: A test of enemy-free space. *Ecology* 69: 1506-1516.

Hindayana, D., R. Meyhofer, D. Scholz & H.M. Poehling. 2000. Intraguild predation

among the hoverfly *Episyrphus balteatus* de Geer (Diptera: Syrphidae) and other aphidophagous predators. *Biol. Control* 20: 236-246.

Kan, E. 1988a. Assessment of aphid colonies by hoverflies. I Maple aphids and *Episyrphus balteatus* (de Geer) (Diptera: Syrphidae). *J. Ethol.* 6: 39-48.

Kan, E. 1988b. Assessment of aphid colonies by hoverfly. II Pea aphids and 3 syrphid species; *Betasyrphus serarius* (Wiedemann), *Metasyrphus frequens* Matsumura and *Syrphus vitripennis* (Meigen) (Diptera: Syrphidae). *J. Ethol.* 6: 135-142.

Kan, E. & M. Sasakawa. 1986. Assessment of the maple aphid colony by the hover fly *Episyrphus balteatus* (de Geer) (Diptera: Syrphidae) I. *J. Ethol.* 4: 121-127.

Kanno, H. & M.O. Harris. 2002. Avoidance of occupied hosts by the Hessian fly: oviposition behaviour and consequences for larval survival. *Ecol. Entomol.* 27: 177-188.

Kiflawi, M., L. Blaustein & M. Mangel. 2000. Oviposition habitat selection by the mosquito *Culiseta longiareolata* in response to risk of predation and conspecific larval density. *Ecol. Entomol.* 28: 168-173.

Lucas, E., D. Coderre & J. Brodeur. 1999. Intraguild predation among aphid predators: characteristics and influence of extraguild predator density. *Ecology* 79: 1084-1092.

Meyhofer, R. & D. Hindayana. 2000. Effect of intraguild predation on aphid parasitoid survival. *Entomol. Exp. Appl.* 97: 115-122.

Mira, A. & E.A. Bernays. 2002. Trade off host use by *Manduca sexta*: plant characteristics vs natural enemies. *Oikos* 97: 387-397.

- Novotny, V., Y. Basset, J. Auga, W. Boen, C. *Entomol. Exp. Appl.* 89: 47-55.
- Sadeghi, H. & F. Gilbert. 2000. Aphid suitability and its relationship to oviposition preference in predatory hoverflies. *J. Anim. Ecol.* 69: 771-784.
- Sato, S. & A.F.G. Dixon. 2004. Effect of intraguild predation on the survival and development of three species of aphidophagous ladybirds: consequences for invasive species. *Agric. For. Entomol.* 1: 21-24.
- Schellhorn, N.A. & D.A. Andow. 1999. Cannibalism and interspecific predation: role of oviposition behavior. *Ecol. Appl.* 9: 418-428.
- Singer, M.S., D. Rodrigues, J.O. Stireman & Y. Carriere. 2004. Roles of food quality and enemy-free space in host use by a generalist insect herbivore. *Ecology* 85: 2747-2753.
- Snyder, W.E. & D.H. Wise (1999) Predator interference and the establishment of generalist predator populations for biocontrol. *Biol. Control* 15: 283-292.
- Sokal, R.R. & F.J. Rohlf. 1995. *Biometry*. Freeman and Company, New York.
- Takizawa, T., H. Yasuda & B.K. Agarwala. 2000. Effect of three species of predatory ladybirds on oviposition of aphid parasitoids. *Entomol. Sci.* 3: 465-469.
- Taylor, A.J., C.B. Muller & H.C.J. Godfray. 1998. Effect of aphid predators on oviposition behavior of aphid parasitoids. *J. Insect. Behav.* 11: 297-302.
- Tenhumberg, B. 1995. Estimating predatory efficiency of *Episyrphus balteatus* (Diptera, Syrphidae) in cereal fields. *Environ. Entomol.* 24: 687-691.
- Yasuda, H. & N. Ohnuma. 1999. Effect of cannibalism and predation on the larval performance of two ladybird beetles. *Entomol. Exp. Appl.* 93: 63-67.
- Yasuda, H. & K. Shinya. 1997. Cannibalism and interspecific predation in two predatory ladybirds in relation to prey abundance in the field. *Entomophaga* 42: 153-163.